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Influence of different dietary vitamin C levels on vitamin E and C content and oxidative stability in various tissues and stored *m. longissimus dorsi* of growing pigs

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Abstract

Both vitamin E and C have antioxidative properties and may act synergistically. To examine a possible interaction between vitamin E, C and oxidative stability in various tissues, 40 barrows (25–105 kg body weight) were allocated to four cereal-based diets (13.4 MJ digestible energy/kg, 168 g crude protein/kg, 140 mg dietary DL-α-tocopheryl acetate) which were supplemented with 0 (B), 150 (C100), 300 (C200) or 600 (C400) mg/kg crystalline ascorbic acid. The influence of storage time on these factors in *m. longissimus dorsi* samples (LD) was investigated. Samples of liver (LI), heart (HT), spleen (SP), backfat outer layer (BF) and LD were obtained. Dry matter, vitamin E, vitamin C and thiobarbituric acid reactive substances (TBARS) were analyzed in all investigated tissues and in addition color (L^*, a^*, b^* values) and drip loss measurements in LD at day 0, 1, 2, 3, 4 and 8 were collected. The treatments did not influence growth performance and slaughter data of the pigs. Neither the vitamin E nor the vitamin C content of investigated tissues showed differences due to feeding treatments. There was also a lack of response to the amount in TBARS. Parameters in LD samples were only affected by storage time and not by diets. The results show that dietary use of both vitamins simultaneously did not further improve qualitative characteristics of the investigated pork tissues.

Keywords: Growing pigs; Vitamin E; Vitamin C; Oxidative stability; Tissues; Storage time

1. Introduction

As is well known, vitamin E in the form of α-tocopherol is a powerful antioxidant and widely used in pig nutrition. Its main location of action is in the cell membranes where it protects phospholipids against oxidation. Water-soluble vitamin C acts as an antioxidant in aqueous environments of cellular components. Both vitamins may act synergistically. Ascorbic acid can regenerate tocopheroxyl radicals to vitamin E, which is able to act again as an antioxidant (Packer, Slater, & Willson, 1979) or can be stored. Leonhardt, Gebert, and Wenk (1996) and Eichenberger, Pfirter, Wenk, and Gebert (2004) found that the vitamin E content in pork tissues can be improved by dietary vitamin C.

Color as well as oxidative stability and drip loss, are important factors in pork quality. Supplementation with vitamin E led to lower susceptibility to oxidation of pork meat (Hoving-Bolink, Eikelenboom, van Diepen, Jongbloed, & Houben, 1998; Monahan et al., 1990). In the present study, the influence of different dietary vitamin C levels on vitamin E and C content and oxidative stability, measured as thiobarbituric acid reactive substances (TBARS), in various pork tissues were investigated. Furthermore, meat color and drip loss were determined in *m. longissimus dorsi* samples during storage.

2. Materials and methods

2.1. Experimental design, animals and diets

Forty Large White barrows were allocated to four feeding treatments according to litter origin and initial body

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weight (BW) and fattened from a mean BW of 24–106 kg. The composition of the basal diet (Table 1) corresponded to commonly used diets based on nutritional recommendations for growing pigs (NRC, 1998). Digestible energy (DE) and crude protein (CP) content were calculated to be 13.4 MJ/kg DE and 168 g/kg CP, respectively (Table 2). The animals were fed with either the basal diet (treatment B) or the basal diet supplemented with 150 mg/kg (C100), 300 mg/kg (C200) or 600 mg/kg (C400) vitamin C as crystalline ascorbic acid (Table 1). The diets were pelleted (4.5 mm diameter) at 60 °C. BW was recorded weekly. The animals were kept in individual pens and fed once daily according to a BW-based feeding scale ($190\,g \times BW^{0.569}$) with water available ad libitum. The experimental procedures described, were approved by the official authority of the canton Zug (authorization number ZG 85).

2.2. Sampling procedure

The animals ($n = 39$) were slaughtered at the abattoir of the MLP Sempach (Swiss Pig Performance Testing Station, Sempach, Switzerland). After bleeding, samples of liver, spleen and heart were obtained within 15 min. Samples for vitamin C determination were separated immediately, frozen in liquid nitrogen, vacuum-packed and kept at −20 °C. For the other analyses the samples were cooled until further processing within 2 h. The organ samples were trimmed of extramuscular tissues, homogenized (Moulinette, type SE, Moulinex) and stored for −20 °C for 8 weeks.

Carcass dissection procedures and measuring of meat quality parameters were carried out according to MLP cutting standards (Rebsamen, Schwörer, & Lorenz, 1995). Carcasses were split centrally and chilled at 0 °C for 24 h prior to dissection of the left carcass side. Loin samples from 6th to 14th rib were obtained, *m. longissimus dorsi* was separated and from the 9th rib, two pieces (2 cm thick) were cut for color and drip loss measurement. The backfat along the split line (width 5 cm) was removed from each animal. The rest of *m. longissimus dorsi* samples and separated backfat outer layer were treated in the same way as the organ samples.

2.3. Analytical methods

The feed content of dry matter (DM), crude ash, crude fiber, crude lipids and CP ($6.25 \times N$) were determined by the standard procedure of VDLUFA (Naumann, Bassler, Seibold, & Barth, 1997). Gross energy (GE) was assessed with an anisothermic bomb calorimeter (C 700 T, IKA Analysentechnik GmbH, Heitersheim, Germany).

The pH measurements (Winiton® Absolute pH-meter, 10 pHW 16, Gerzensee, Switzerland) of meat were carried out with a cute-in electrode (Ingold by Mettler-Toledo AG, Urdorf, Switzerland) 45 min (pH 1) and 24 h (pH 2) Postmortem (p.m.) behind the 10th rib (*m. longissimus dorsi*) at the left side of carcass. The determination of DM in various tissues was performed analogous to the feed samples.

For the vitamin E analyses, thawed tissue samples were analyzed as described by Rettenmaier and Schüep (1992) using HPLC. The content of vitamin C was analyzed by HPLC at the laboratories of F. Hoffmann-La Roche Ltd., Basel (Drouard, 2001).

TBARS were determined in samples of backfat outer layer, liver, spleen and *m. longissimus dorsi* by the distillation method of Tarladgis, Watts, Younathan, and Dugan (1960), with certain modifications as described by Kunz and Prabucki (1986).

The oxidative stability of backfat outer layer was also measured by means of the Rancimat test (Ranzimat 679, Metrohm AG, Herisau, Switzerland) based on Pardun and Kroll (1972). Fat samples were melted at 90 °C during 3 h and the resulting lipid fraction was kept under vacuum

Table 1
Experimental design and composition of the basal diet

	Feeding treatment			
Item	B	C100	C200	C400
Number of animals	10	10	10	10
Supplement (mg/kg)				
Vitamin C[a]	–	150	300	600
Basal diet (%)				
Barley	46.4	Limestone meal		0.64
Wheat	19.8	Dicalcium phosphate		1.2
Soybean meal 43	17.9	Salt		0.33
Potato meal	5.0	Celite 545 (Indicator)		1.2
Lysine–HCl	0.46	Beef/pork tallow 65		2.5
Methionine 20%	0.77	Molasse		2.5
Threonine 20%	0.8	Vitamin/mineral premix[b]		0.50

[a] Crystalline ascorbic acid from DSM Nutritional Products Ltd., Basel, added in 150% rate of the expected amount (100, 200, 400 mg kg^{-1}).

[b] Supplied the following per kilogram of diet: vitamin A, 10000 IU; vitamin D_3, 1000 IU; vitamin E, 140 IU; vitamin B_2, 4 mg; vitamin B_6, 4 mg; vitamin B_{12}, 0.015 mg; vitamin K_3, 0.1 mg; pantothenic acid, 15 mg; niacin, 20 mg; folic acid, 0.2 mg; Fe, 60 mg ($FeSO_4$); I, 1 mg ($Ca(IO)_3$); Se, 0.3 mg (Na_2Se); Cu, 15 mg ($CuSO_4$); Zn, 100 mg (ZnO_2); Mn, 40 mg (MnO_2).